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Short Editorial Review

Endothelin in renal disease: role of endothelin antagonists

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In spite of tremendous progress in the understanding of human disease, the search for new mediators continues as numerous conditions remain poorly understood. In particular in many forms of renal disease, effective and cause-orientated forms of therapy are still lacking. The discovery of new pathogenetic mechanisms often leads to the development of new drugs with previously unknown properties which may offer new treatment modalities. Endothelin is a recently discovered potent biological mediator which exists in three closely related isoforms (i.e. endothelin-1, -2 and -3 [1–3]. Endothelins belong to a family of 21 amino acid peptides with two disulphide bridges [1,3]. The main vascular effects of endothelin are transient vasodilatation and profound and sustained vasoconstriction [4]. Endothelin also exerts marked renal effects, acts as a mitogen and stimulates proliferation of vascular smooth muscle and glomerular mesangial cells [5–7].

Important sources of the peptides are endothelial cells, neurons, renal cells and at least under certain conditions also vascular smooth muscle cells. Stimuli for the release of endothelins include hypoxia and/or ischaemia [8], but also humoral factors (angiotensin II, vasopressin, transforming growth factor-beta, insulin, thrombin and several cytokines) and potentially nephrotoxic drugs such as radiocontrast agents, cyclosporin, amphotericin B and OKT-3 [1,3,9,10–22]. On the other hand, nitric oxide and atrial natriuretic peptide inhibit endothelin production via a cyclic GMP-dependent mechanism [1,3,11,20]. In addition, smooth muscle cells appear to release an inhibitory factor which limits the production of the peptide. This may explain why intact tissues such as the blood vessel wall produce markedly less endothelin than isolated cells in culture. *In vivo* in humans, endothelin plasma levels are very low [21]. However, in different disease states, elevated endothelin plasma levels have been described (see below, Table 1).

In the kidney, endothelin reduces renal blood flow and glomerular filtration rate [10,22–25]; this is mainly due to vasoconstriction of both afferent and efferent arterioles. Systemic infusion of endothelin-1 in humans *in vivo* leads to blood pressure increase, sodium retention and reduction in urine flow [24,25]. Although *in vitro* endothelin inhibits renin release [23,26], in the intact organism renin plasma levels do not change or increase (due to renal vasoconstriction) after infusion of endothelin-1 [25]. Endothelin stimulates release of aldosteron, vasopressin and atrial natriuretic peptide under experimental conditions [9,10]. However, *in vivo* in humans, it does not influence the plasma levels of these hormones [24]. The role of the mitogenic properties of endothelin [5–7] in the kidney is still unclear, but it could be involved in proliferative glomerular diseases.

Endothelins exert their biological effects via activation of specific receptors. These membrane-bound receptors have seven transmembrane domains and are coupled to G-proteins; three types of endothelin receptors have been cloned, i.e. ET_A, ET_B and ET_C receptors [27]. Endothelin-1, the primary product of endothelial cells, preferentially activates ET_A receptors. ET_B receptors exert no isoform specificity and are equally activated by all endothelin isoforms, while the ET_C receptor preferentially binds endothelin-3 [28]. ET_A receptors on vascular smooth muscle cause vasoconstriction and mediate proliferation, although ET_B receptors contribute to these effects. Endothelial cells express only ET_B receptors linked to nitric oxide and prostacyclin formation. The endothelin receptors on renal cells have only partially been characterized. However, the ET_A receptors seem to be expressed mainly in the glomerulus, the vasa recta bundle and the arcuate artery, while the ET_B receptor predominates in the initial and terminal

Table 1. Possible role of endothelin in disease

Heart disease	Myocardial infarction
	Coronary spasm
	Cardiac shock
	Heart failure
Vascular disease	Atherosclerosis
	Takayashu's disease
	Raynaud's disease
Hypertension	Arterial hypertension (?)
	High altitude pulmonary hypertension
Other	Migraine
	Subarachnoid haemorrhage
	Renal failure
	Hepatorenal syndrome
	Low tension glaucoma

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inner medullary collecting duct and also in the glomerulus [29].

Recently, new molecules have been synthesized, which *in vitro* inhibit the effects of endothelin (Table 2) [30–34]. Certain of these molecules inhibit ET_A receptors only, while others interfere with both ET_A and ET_B receptors. As both ET_A and ET_B receptors are expressed on vascular smooth muscle also in the human [35–37], combined antagonists more effectively interfere with the vasoconstrictor effects of endothelin. On the other hand, non-selective endothelin antagonists also reduce the release of nitric oxide and prostacyclin from the endothelium and hence the potentially beneficial vasodilator effects of endothelin. In the human skin microcirculation *in vivo*, both combined and selective endothelin antagonists potently inhibit the vasoconstrictor effects of endothelin-1 [38]. In the kidney, it appears that both receptors contribute to the effects of endothelin. However, although both ET_A and ET_B receptors are present, their distribution in the kidney is not uniform suggesting different function [29]. Hence, it is likely that combined endothelin antagonists are required to interfere with the renal effects of endothelin.

Several experimental studies suggest a pathophysiologic role of endothelin in renal failure. Indeed, acute ischaemic renal failure leads to an increase in endothelin release and/or endothelin receptor upregulation [39–42]; this effect can be reversed by endothelin receptor antagonists or drugs blocking the endothelin converting enzyme [43–45]. In uraemic patients, endothelin plasma levels are elevated [46]. In particular, renal failure induced by nephrotoxic agents seems to be related to increased endothelin levels; this is true for radiocontrast agents independent of their type [47–50], as well as for other potentially nephrotoxic drugs, like amphotericin B [51] and possibly cisplatin [52]. Immunosuppressive agents such as cyclosporin and FK 506 also modulate endothelin release (see below). Obviously, acute renal failure can be caused by a variety of stimuli and the importance of a given mediator may vary depending on the major cause involved. Ischaemic renal failure is relatively well defined experimentally and is particularly suitable for studies with newly developed drugs. Indeed, in a monkey model, decreases in renal blood flow in acute ischaemic renal failure could be prevented by an ET_A/ET_B receptor antagonist [53].

Table 2. Endothelin antagonists

Drug	Receptor
BE-18257A/B	ET_A
BQ-162	ET_A
BQ-123	ET_A
BQ-153	ET_A
PD 147953 (=FR 139317)	ET_A
RO-462005	ET_A/ET_B
PD 142893	ET_A/ET_B
PD 145065	ET_A/ET_B

In chronic renal failure, only very few studies exist; they all found increases in endothelin plasma levels and/or gene expression [54,55]. The mechanisms involved are not entirely clear. Endothelin levels could be increased because of (1) a reduced renal clearance, (2) loss of inhibitory mechanisms in renal failure or (3) due to stimulatory effects, i.e. of uraemic toxins. In patients with renal failure an endogenous inhibitor of the l-arginine/nitric oxide pathway (i.e. dimethylarginine or ADMA [55] accumulates. The inhibition of nitric oxide production could explain an increased endothelin production as nitric oxide reduces endothelin production from the blood vessel wall via a cGMP-dependent mechanism [11]. Patients on haemodialysis also have elevated plasma endothelin levels [46]. The volume contraction after haemodialysis seems to increase plasma levels of endothelin further, possibly through an activation of the sympathetic nervous system [46,57,58], which is known to increase plasma endothelin levels [59,60]. This may explain the differences observed between peritoneal dialysis (CAPD) and no dialysis on the one hand and haemodialysis on the other, although this is controversial [46,58]. High-flux membranes (PAN, PMMA, CTA) seem to clear endothelin better than normal membranes [61]. Whether these findings reflect a pathogenetic role for endothelin in chronic renal disease in humans will be clarified by appropriate clinical studies with endothelin antagonists in the future.

In disease states with secondary involvement of the kidney—like diabetes [62], hepatorenal syndrome [63], thrombotic thrombopenic purpura [64], septic shock [65,66], but also in congestive heart failure [67–70] and severe atherosclerosis [71]—elevated plasma levels of endothelin have been described and may contribute to the deterioration of renal function occurring under these conditions. Ongoing experimental and clinical trials with endothelin antagonists will elucidate the pathogenic role of endothelins in these disease states and its involvement in renal functional impairment in particular. Similarly, in autoimmune diseases like Morbus Wegener and Morbus Raynaud [59], elevated endothelin plasma levels occur. In a mice lupus nephritis model, endothelin gene expression is increased and tends to normalize with prednisolone therapy [72]. It is conceivable, therefore, that inflammatory diseases of the vascular wall or the glomerulus are associated with an activation of the endothelin axis. Whether or not such an activation is primary or secondary in nature in these diseases requires experimental and later clinical studies with specific endothelin receptor antagonists.

The role of endothelin in arterial hypertension is controversial (see [2,73]). Infusion of endothelin does increase blood pressure in experimental animals and in humans [10,74]. Moreover, patients with endothelin-secreting haemangioendotheliomas are hypertensive [75]. Whether or not endothelin production is altered in hypertension is uncertain. Although some studies found increased plasma levels of endothelin, many other studies found no differences as compared to

controls. However, circulating endothelin may not reflect local levels of the peptide, as in the blood vessel wall endothelin is primarily released abluminally [76]. Indeed, in DOCA-salt hypertension vascular endothelin production is increased in the presence of normal plasma levels of the peptide [77]. In the SHR, however, both circulating and vascular endothelin is suppressed. Similarly, in the renal medulla of the SHR, the endothelin content is reduced [78]. These experimental findings suggest that endothelin may be differently involved in different forms of hypertension, possibly also in the human. To further elucidate the role of endothelin, transgenic and gene knockout rats have been produced. Endothelin-2 transgenic rats are normotensive (possibly because of the activation of compensatory vasodilator mechanisms) and endothelin-1 gene knock-out mice are actually hypertensive [79]. For the interpretation of the data derived from the latter models, one has to be reminded of the fact that in humans increased endothelin levels, for instance derived from vascular tumours, do indeed cause hypertension. Moreover, in the human hand vein circulation of patients with essential hypertension, the vasoconstrictor response to endothelin is increased. Finally, endothelin antagonists lower blood pressure in salt-depleted monkeys in the SHR and DOCA-salt hypertensive rats. The surprising finding that endothelin knock-out rats have profound malformations of the throat indicates that the peptide may be importantly involved in the development of these organs [79].

Cyclosporin therapy is established in the treatment of host versus graft rejection. However, the drug often leads to hypertension and impairs renal function. In cultured endothelial cells cyclosporin stimulates endothelin production [80,81]. Furthermore, in the renal medulla of rats [82] and rabbits [83], cyclosporin, and even more so its metabolites [83], but also FK 506 [84], stimulate the production of endothelin, inhibit prostacyclin release and in turn lead to renal vasoconstriction, especially in the afferent arteriole [85]. In addition, both cyclosporin and FK 506 have cytotoxic effects in renal cells [84]. In transplant recipients, cyclosporin increases endothelin levels [86]. In the rat renal circulation, cyclosporin reduces renal blood flow; this effect can be prevented or reversed by endothelin antibodies or endothelin antagonists [87]. However, whether inhibition of endothelin can prevent cyclosporin-induced side effects, especially nephrotoxicity and hypertension, has still to be established.

Hence, in summary, the endothelins are a new and potentially very important family of peptides with potent effects in the cardiovascular system and the kidney in particular. Their biological effects could explain a variety of disturbances occurring in renal disease and in several forms of hypertension. Of great interest for renal physiologists and nephrologists is the fact that the endothelin system is activated in several renal diseases and that endothelin antagonists are effective in reversing impaired renal function in experimental models. Definitive proof for an involvement of endothelins in renal function and disease in humans awaits the results of ongoing clinical trials with new and specific endothelin receptor antagonists.

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References

1. Lüscher TF, Boulanger CM, Dohi Y, Yang ZH. Endothelium-derived contracting factors. *Hypertension* 1992; 19: 117–130.
2. Lüscher TF, Seo BG, Bühler FR. Potential role of endothelin in hypertension: controversy of endothelin in hypertension. *Hypertension* 1993; 21: 752–757.
3. Yanagisawa M, Kurihara H, Kimura S, Tomobe Y, Kobayashi M, Mitsui Y, Yazaki Y, Goto K, Masaki T. A novel potent vasoconstrictor peptide produced by vascular endothelial cells. *Nature* 1988; 332: 411–415.
4. Kiowski W, Lüscher TF, Linder L, Buhler FR. Endothelin-1-induced vasoconstriction in humans. Reversal by calcium channel blockade, but not by nitrovasodilators or endothelium-derived relaxing factor. *Circulation* 1991; 83: 469–475.
5. Simonson MS, Wann S, Mene P, Dubyak GR, Kester M, Nakazato Y, Sedor JR, Dunn MJ. Endothelin stimulates phospholipase C, Na^+/H^+ exchange, c-fos expression, and mitogenesis in rat mesangial cells. *J Clin Invest* 1989; 83: 708–712.
6. Takuwa N, Takuwa Y, Yanagisawa M, Yamashita K, Masaki T. A novel vasoactive peptide endothelin stimulates mitogenesis through inositol lipid turnover in Swiss 3T3 fibroblasts. *J Biol Chem* 1989; 264: 7856–7861.
7. Hirata Y, Takagi Y, Fukuda Y, Marumo F. Endothelin is a potent mitrogen for rat vascular smooth muscle cells. *Atherosclerosis* 1989; 78: 225–228.
8. Rakugi H, Tabuchi Y, Nakamaru M, Nagano M, Higashimori K, Mikami H, Ogihara T, Suzuki N. Evidence for endothelin-1 release from resistance vessels of rats in response to hypoxia. *Biochem Biophys Res Commun* 1990; 169: 973–977.
9. Rosolowsky LJ, Campbell WB. Endothelin enhances adrenocorticotropin-stimulated aldosterone release from cultured bovine adrenal cells. *Endocrinology* 1990; 126: 1860–1866.
10. Goetz KL, Wang BC, Madwed JB, Zhu JL, Leadley R Jr. Cardiovascular, renal, and endocrine responses to intravenous endothelin in conscious dogs. *Am J Physiol* 1988; 255: R1064–R1068.
11. Boulanger C, Lüscher TF. Release of endothelin from the porcine aorta. Inhibition of endothelium-derived nitric oxide. *J Clin Invest* 1990; 85: 587–590.
12. Yanagisawa M, Inoue A, Ishikawa T, Kasuya Y, Kimura S, Kumagaye S, Nakajima K, Watanabe TX, Sakakibara S, Goto K, Masaki T. Primary structure, synthesis, and biological activity of rat endothelin, an endothelium-derived vasoconstrictor peptide. *Proc Natl Acad Sci USA* 1988; 85: 6964–6967.
13. Dohi Y, Hahn AW, Boulanger CM, Bühler FR, Lüscher TF. Endothelin stimulated by angiotensin II augments contractility of spontaneously hypertensive rat resistance arteries. *Hypertension* 1992; 19: 131–137.
14. Emori T, Hirata Y, Ohta K, Kanno K, Eguchi S, Imai T, Shichiri M, Marumo F. Cellular mechanism of endothelin-1 release by angiotensin and vasopressin. *Hypertension* 1991; 18: 165–170.
15. Hattori Y, Kasai K, Nakamura T, Emoto T, Shimoda S. Effect of glucose and insulin on immunoreactive endothelin-1 release from cultured porcine aortic endothelial cells. *Metabolism* 1991; 40: 165–169.
16. Endo T, Uchida Y, Matsumoto H, Suzuki N, Nomura A, Hirata F, Hasegawa S. Regulation of endothelin-1 synthesis in cultured guinea pig airway epithelial cells by various cytokines. *Biochem Biophys Res Commun* 1992; 186: 1594–1599.
17. Kohno M, Yasunari K, Murakawa K, Yokokawa K, Horio T,

Fukui T, Takeda T. Release of immunoreactive endothelin from porcine aortic strips. *Hypertension* 1990; 15: 718–723.
18. Yoshizumi M, Kurihara H, Morita T, Yamashita T, Oh-Hashi Y, Sugiyama T, Takaku F, Yanagisawa M, Masaki T, Yazaki Y. Interleukin 1 increases the production of endothelin-1 by cultured endothelial cells. *Biochem Biophys Res Commun* 1990; 166: 324–329.
19. Yamashita J, Ogawa M, Nomura K, Matsuo S, Inada K, Yamashita S, Nakashima Y, Saishoji T, Takano S, Fujita S. Interleukin 6 stimulates the production of immunoreactive endothelin 1 in human breast cancer cells. *Cancer Res* 1993; 53: 464–467.
20. Saijonmaa O, Ristimaki A, Fyhrquist F. Atrial natriuretic peptide, nitroglycerine, and nitroprusside reduce basal and stimulated endothelin production from cultured endothelial cells. *Biochem Biophys Res Commun* 1990; 173: 514–520.
21. Suzuki N, Matsumoto H, Kitada C, Yanagisawa M, Miyauchi T, Masaki T, Fujino M. Immunoreactive endothelin-1 in plasma detected by a sandwich-type enzyme immunoassay. *J Cardiovasc Pharmacol* 1989; 13 (Suppl 5): 151–152.
22. Bloom IT, Bentley FR, Wilson MA, Garrison RN. *In vivo* effects of endothelin on the renal microcirculation. *J Surg Res* 1993; 54: 274–280.
23. Lin H, Sangmal M, Smith M Jr, Young DB. Effect of endothelin-1 on glomerular hydraulic pressure and renin release in dogs. *Hypertension* 1993; 21: 845–851.
24. Sorensen SS, Madsen JK, Pedersen EB. Systemic and renal effect of intravenous infusion of endothelin-1 in healthy human volunteers. *Am J Physiol* 1994; 266: F411–F418.
25. Rabelink TJ, Kaasjager KAH, Boer P, Stroes EG, Braam B, Koomans HA. Effects of endothelin-1 on renal function in humans: Implications for physiology and pathophysiology. *Kidney Int* 1994; 46
26. Naess PA, Christensen G, Kill F. Inhibitory effect of endothelin on renin release in dog kidneys. *Acta Physiol Scand* 1993; 148: 131–136.
27. Kurihara T, Akimoto M, Kurokawa K, Ishiguro H, Niimi A, Maeda A, Sigemoto M, Yamashita K, Yokoyama I, Hirayama Y, *et al.* ET-3 sensitive reduction of tissue blood flow in rat liver. *Life Sci* 1992; 51: 6.
28. Karne S, Jayawickreme CK, Lerner MR. Cloning and characterization of an endothelin-3 specific receptor (ETC receptor) from *Xenopus laevis* dermal melanophores. *J Biol Chem* 1993; 268: 19126–19133.
29. Terada Y, Tomita K, Nonoguchi H, Marumo F. Different localization of two types of endothelin receptor mRNA in microdissected rat nephron segments using reverse transcription and polymerase chain reaction assay. *J Clin Invest* 1992; 90: 107–112.
30. Aramori I, Nirei H, Shoubo M, Sogabe K, Nakamura K, Kojo H, Notsu Y, Ono T, Nakanishi S. Subtype selectivity of a novel endothelin antagonist, FR139317, for the two endothelin receptors in transfected Chinese hamster ovary cells. *Mol Pharmacol* 1993; 43: 127–131.
31. Cardell LO, Uddman R, Edvinsson L. A novel ETA-receptor antagonist, FR 139317, inhibits endothelin-induced contractions of guinea-pig pulmonary arteries, but not trachea. *Br J Pharmacol* 1993; 108: 448–452.
32. Sogabe K, Nirei H, Shoubo M, Nomoto A, Ao S, Notsu Y, Ono T. Pharmacological profile of FR139317, a novel, potent endothelin ETA receptor antagaonist. *J Pharmacol Exp Ther* 1993; 264: 1040–1046.
33. Eguchi S, Hirata Y, Ihara M, Yano M, Marumo F. A novel ETA antagonist (BQ-123) inhibits endothelin-1-induced phosphoinositide breakdown and DNA synthesis in rat vascular smooth muscle cells. *FEBS Lett* 1992; 302: 243–246.
34. Breu V, Loffler BM, Clozel M. *In vitro* characterization of Ro 46-2005, a novel synthetic non-peptide endothelin antagonist of ETA and ETB receptors. *FEBS Lett* 1993; 334: 210–214.
35. Cristol JP, Warner TD, Thiemermann C, Vane JR. Mediation via different receptors of the vasoconstrictor effects of endothelins and sarafotoxins in the systemic circulation and renal vasculature of the anaesthetized rat. *Br J Pharmacol* 1993; 108: 776–779.
36. Pollock DM, Opgenorth TI. Evidence for endothelin-induced renal vasoconstriction independent of ETA receptor activation. *Am J Physiol* 1993; 264: R222–R226.
37. Seo BG, Siebenmann R, Lüscher TF. ETA- and ETB-receptors mediate contractions in human blood vessels. *Hypertension* 1993; 22: 425.
38. Wenzel RR, Noll G, Lüscher TF. Endothelin receptor antagonists inhibit endothelin in human skin microcirculation. *Hypertension* 1994; 23: 581–586.
39. Roubert P, Cornet S, Plas P, Guilmard C , Pirotzky E, Chabrier PE, Braquet P. Upregulation of renal endothelin receptors in glycerol-induced acute renal failure in the rat. *J Cardiovasc Pharmacol* 1993; 22: S303–S305.
40. Tomita K, Ujiie K, Nakanishi T, Tomura S, Matsuda O, Ando K, Shichiri M, Hirata Y, Marumo F. Plasma endothelin levels in patients with acute renal failure (letter). *N Engl J Med* 1989; 321: 16.
41. Warrens AN, Cassidy MJ, Takahashi K, Ghatei MA, Bloom SR. Endothelin in renal failure. *Nephrol Dial Transplant* 1990; 5: 418–422.
42. Nambi P, Pullen M, Jugus M, Gellai M. Rat kidney endothelin receptors in ischemia-induced acute renal failure. *J Pharmacol Exp Ther* 1993; 264: 345–348.
43. Vemulapalli S, Chiu PJ, Chintala M, Bernardino V. Attenuation of ischemic acute renal failure by phosphoramidon in rats. *Pharmacology* 1993; 47: 188–193.
44. Mino N, Kobayashi M, Nakajima A, Amano H, Shimamoto K, Ishikawa K, Watanabe K, Nishikibe M, Yano M, Ikemoto F. Protective effect of a selective endothelin receptor antagonist, BQ-123, in ischemic acute renal failure in rats. *Eur J Pharmacol* 1992; 221: 77–83.
45. Gellai M, Jugus M, Fletcher T, De Wolf R, Nambi P. Reversal of postischemic acute renal failure with a selective endothelinA receptor antagonist in the rat. *J Clin Invest* 1994; 93: 900–906.
46. Mallamaci F, Parlongo S, Zoccali C. Influence of cardiovascular damage and residual renal function on plasma endothelin in chronic renal failure. *Nephron* 1993; 63: 291–295.
47. Heyman SN, Clark BA, Cantley L, Spokes K, Rosen S, Brezis M, Epstein FH. Effects of ioversol versus iothalamate on endothelin release and radiocontrast nephropathy. *Invest Radiol* 1993; 28: 313–318.
48. Heyman SN, Clark BA, Kaiser N, Spokes K, Rosen S, Brezis M, Epstein FH. Radiocontrast agents induce endothelin release *in vivo* and *in vitro*. *J Am Soc Nephrol* 1992; 3: 58–65.
49. Cantley LG, Spokes K, Clark B, McMahon EG, Carter J, Epstein FH. Role of endothelin and prostaglandins in radiocontrast-induced renal artery constriction. *Kidney Int* 1993; 44: 217–1223.
50. Margulies KB, Hildebrand FL, Heublein DM, Burnett J Jr. Radiocontrast increases plasma and urinary endothelin. *J Am Soc Nephrol* 1991; 2: 1041–1045.
51. Heyman SN, Clark BA, Kaiser N, Epstein FH, Spokes K, Rosen S, Brezis M. *In vivo* and *in vitro* studies on the effect of amphotericin B on endothelin release. *J Antimicrob Chemother* 1992; 29: 69–77.
52. Ohta K, Hirata Y, Shichiri M, Ichioka M, Kubota T, Marumo F. Cisplatin-induced urinary endothelin excretion (letter) *J Am Med Assoc* 1991; 265: 1391–1392.
53. Clozel M, Breu V, Burri K, Cassal JM, Fischli W, Gray GA, Hirth G, Loffler BM, Muller M, Neidhart W, *et al.* Pathophysiological role of endothelin revealed by the first orally active endothelin receptor antagonist. *Nature* 1993; 365: 759–761.
54. Orisio S, Benigni A, Bruzzi I, Corna D, Perico N, Zoja C, Bertani L, Remuzzi G. Renal endothelin gene expression is increased in remnant kidney and correlates with disease progression. *Kidney Int* 1993; 43: 354–358.
55. Benigni A, Zoja C, Corna D, Orisio S, Longaretti L, Bertani T, Remuzzi G. A specific endothelin subtype. A receptor antagonist protects against injury in renal disease progression. *Kidney Int* 1993; 44: 440–444.
56. Vallance P, Leone A, Calver A, Collier J, Moncada S. Accumulation of an endogenous inhibitor of nitric oxide synthesis in chronic renal failure. *Lancet* 1992; 339: 572–575.

57. Ross RD, Kalidindi V, Vincent JA, Kassab J, Dabbagh S, Hsu JM, Pinsky WW. Acute changes in endothelin-1 after hemodialysis for chronic renal failure. *J Pediatr* 1993; 122: S74–S76.
58. Deray G, Carayon A, Maistre G, Benhmida M, Masson F, Barthelemy C, Petitclerc T, Jacobs C. Endothelin in chronic renal failure. *Nephrol Dial Transplant* 1992; 7: 300–305.
59. Zamora MR, O'Brien RF, Rutherford RB, Weil JV. Serum endothelin-1 concentrations and cold provocation in primary Raynaud's phenomenon (see comments). *Lancet* 1990; 336: 1144–1147.
60. Fyhrquist F, Saijonmaa O, Metsarinne K, Tikkanen I, Rosenlof K, Tikkanen T. Raised plasma endothelin-I concentration following cold pressor test. *Biochem Biophys Res Commun* 1990; 169: 217–221.
61. Niwa T, Fujishiro T, Uema K, Tsuzuki T, Tominaga Y, Emoto Y, Miyazaki T, Maeda K. Effect of hemodialysis on plasma levels of vasoactive peptides: endothelin, calcitonin gene-related peptide and human atrial natriuretic peptide. *Nephron* 1993; 64: 552–559.
62. Collier A, Leach JP, McLellan A, Jardine A, Morton JJ, Small M. Plasma endothelinlike immunoreactivity levels in IDDM patients with microalbuminuria. *Diabetes Care* 1992; 15: 1038–1040.
63. Moore K, Wendon J, Frazer M, Karani J, Williams R, Badr K. Plasma endothelin immunoreactivity in liver disease and the hepatorenal syndrome [see comments]. *N Engl J Med* 1992; 327: 1774–1778.
64. Spooren PF, Vermes I, Kip L, Haanen C. Endothelin: a possible role in the occurrence of renal failure in thrombotic thrombocytopenic purpura [letter]. *Thromb Haemost* 1993; 69: 401–432.
65. Morise Z, Ueda M, Aiura K, Endo M, Kitajima M. Pathophysiologic role of endothelin-1 in renal function in rats with endotoxin shock. *Surgery* 1994; 115: 199–204.
66. Voerman HJ, Stehouwer CD, van Kamp GJ. Strack van Schijindel RJ, Groeneveld AB, Thijs LG. Plasma endothelin levels are increased during septic shock. *Crit Care Med* 1992; 20: 1097–1101.
67. Margulies KB, Hildebrand FJ, Lerman A, Perrella MA, Burnett JJ. Increased endothelin in experimental heart failure. *Circulation* 1990; 82: 2226–2230.
68. Rodeheffer RJ, Lerman A, Heublein DM, Burnett JJ. Increased plasma concentrations of endothelin in congestive heart failure in humans [see comments]. *Mayo Clin Proc* 1992; 67: 719–724.
69. Stewart DJ, Cernacek P, Costello KB, Rouleau JL. Elevated endothelin-1 in heart failure and loss of normal response to postural change. *Circulation* 1992; 85: 510–517.
70. Cavero PG, Miller WL, Heublein DM, Margulies KB, Burnett JJ. Endothelin in experimental congestive heart failure in the anesthetized dog. *Am J Physiol Renal Fluid Electrolyte Physiol* 1990; 259: 28–32.
71. Lerman A, Edwards BS, Hallett JW, Heublein DM, Sandberg SM, Burnett JJ. Circulating and tissue endothelin immunoreactivity in advanced atherosclerosis. *N Engl J Med* 1991; 325: 997–1001.
72. Nakamura T, Ebihara I, Fukui M, Osada S, Tomino Y, Masaki T, Goto K, Furuichi Y, Koide H. Renal expression of mRNAs for endothelin-1, endothelin-3 and endothelin receptors in NZB/W F1 mice. *Renal Physiol Biochem* 1993; 16: 233–243.
73. Vanhoutte PM. Is endothelin involved in the pathogenesis of hypertension? *Hypertension* 1993; 21: 747–751.
74. Vierhapper H, Wagner OF, Nowotny P, Waldhausl W. Effect of endothelin-1 in man: pretreatment with nifedipine, with indomethacin and with cyclosporine A. *Eur J Clin Invest* 1992; 22: 55–59.
75. Yokokawa K, Tahara H, Kohno M, Murakawa K, Yasunari K, Nakagawa K, Hamada T, Otani S, Yanagisawa M, Takeda T. Hypertension associated with endothelin-secreting malignant hemangioendothelioma. *Ann Intern Med* 1991; 14: 213–215.
76. Wagner OF, Christ G, Wojta J, Vierhapper H, Parzer S, Nowotny PJ, Schneider B, Waldhausl W, Binder BR. Polar secretion of endothelin-1 by cultured endothelial cells. *J Biol Chem* 1992; 267: 16066–16068.
77. Suzuki N, Miyauchi T, Tomobe Y, Matsumoto H, Goto K, Masaki T, Fujino M. Plasma concentrations of endothelin-1 in spontaneously hypertensive rats and DOCA-salt hypertensive rats. *Biochem Biophys Res Commun* 1990; 167: 941–947.
78. Kitamura K, Tanaka T, Kato J, Ogawa T, Eto T, Tanaka K. Immunoreactive endothelin in rat kidney inner medulla: marked decrease in spontaneously hypertensive rats. *Biochem Biophys Res Commun* 1989; 62: 38–44.
79. Kurihara Y, Kurihara H, Suzuki H, Kodama T, Maemura K, Nagai R, Oda H, Kuwaki T, Cao W-H, Kamada N, Jishage K, Ouchi Y, Azuma S, Toyoda Y, Ishikawa T, Kumada M, Yazaki Y. Elevated blood pressure and craniofacial abnormalities in mice deficient in endothelin-1. *Nature* 1994; 368: 703–710.
80. Ong AC, Jowett TP, Scoble JE, O'Shea JA, Varghese Z, Moorhead JF. Effect of cyclosporin A on endothelin synthesis by cultured human renal cortical epithelial cells. *Nephrol Dial Transplant* 1993; 8: 748–753.
81. Bunchman TE, Brookshire CA. Cyclosporine-induced synthesis of endothelin by cultured human endothelial cells. *J Clin Invest* 1991; 88: 310–314.
82. Iwasaki S, Homma T, Kon V. Site specific regulation in the kidney of endothelin and its receptor subtypes by cyclosporine. *Kidney Int* 1994; 45: 592–597.
83. Copeland KR, Yatscoff RW. Comparison of the effects of cyclosporine and its metabolites on the release of prostacyclin and endothelin from mesangial cells. *Transplantation* 1992; 53: 640–645.
84. Moutabarrik A, Ishibashi M, Fukunaga M, Kameoka H, Kawaguchi N, Takano Y, Kokado Y, Sonoda T, Onishi S, Takahara S, *et al.* FK506-induced kidney tubular cell injury. *Transplantation* 1992; 54: 1041–1047.
85. Lanese DM, Conger JD. Effects of endothelin receptor antagonist on cyclosporine-induced vasoconstriction in isolated rat renal arterioles. *J Clin Invest* 1993; 91: 2144–2149.
86. Grieff M, Shoheib SA, Lörtscher R, Stewart DJ. Cyclosporine A induces elevation in circulating endothelin-1 following transplantation. *J Am Soc Nephrol* 1990; 1: 758.
87. Kon V, Awazu M. Endothelin and cyclosporine nephrotoxicity. *Renal Fail* 1992; 14: 345–350.